Fig 1

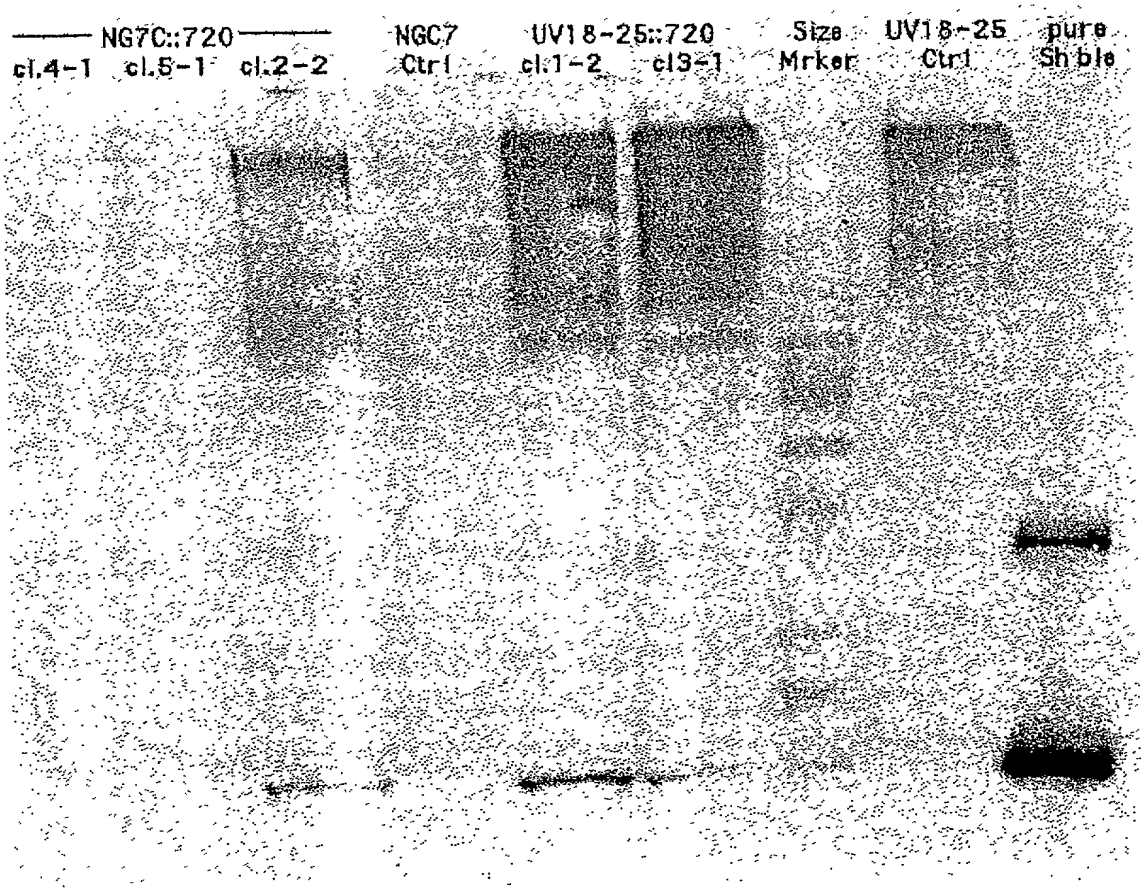

NG7C::720
cl.4-1
cl.5-1
cl.2-2
NGC7
Ctrl
UV18-25::720
cl.1-2
cl3-1
Size
Mrker
UV18-25
Ctrl
pure
Shble

Fig 2

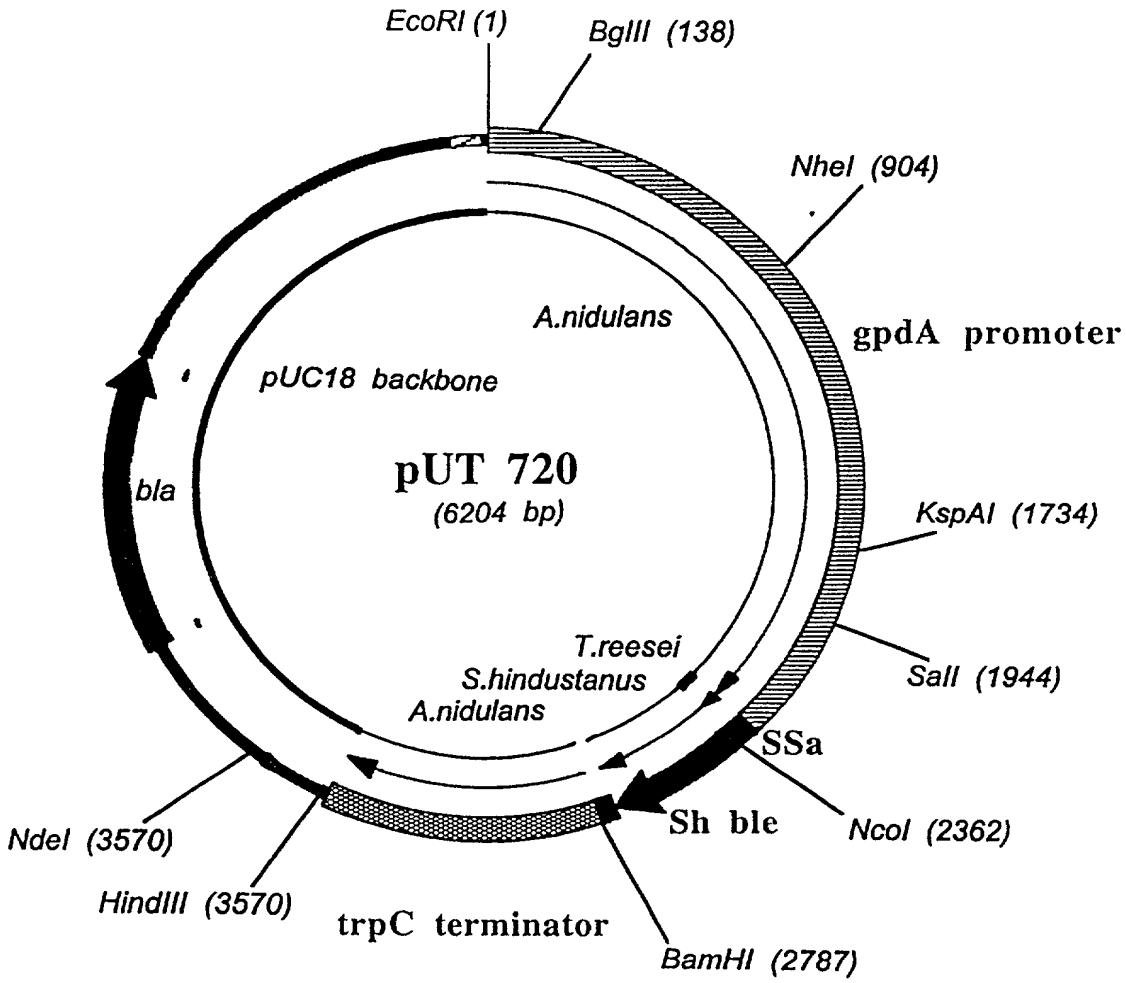

EcoRI (1)
BglII (138)
NheI (904)
gpdA promoter
A.nidulans
pUC18 backbone
pUT 720
(6204 bp)
bla
KspAI (1734)
SalI (1944)
T.reesei
S.hindustanus
A.nidulans
SSa
Sh ble
NcoI (2362)
NdeI (3570)
HindIII (3570)
trpC terminator
BamHI (2787)
200

Fig 3

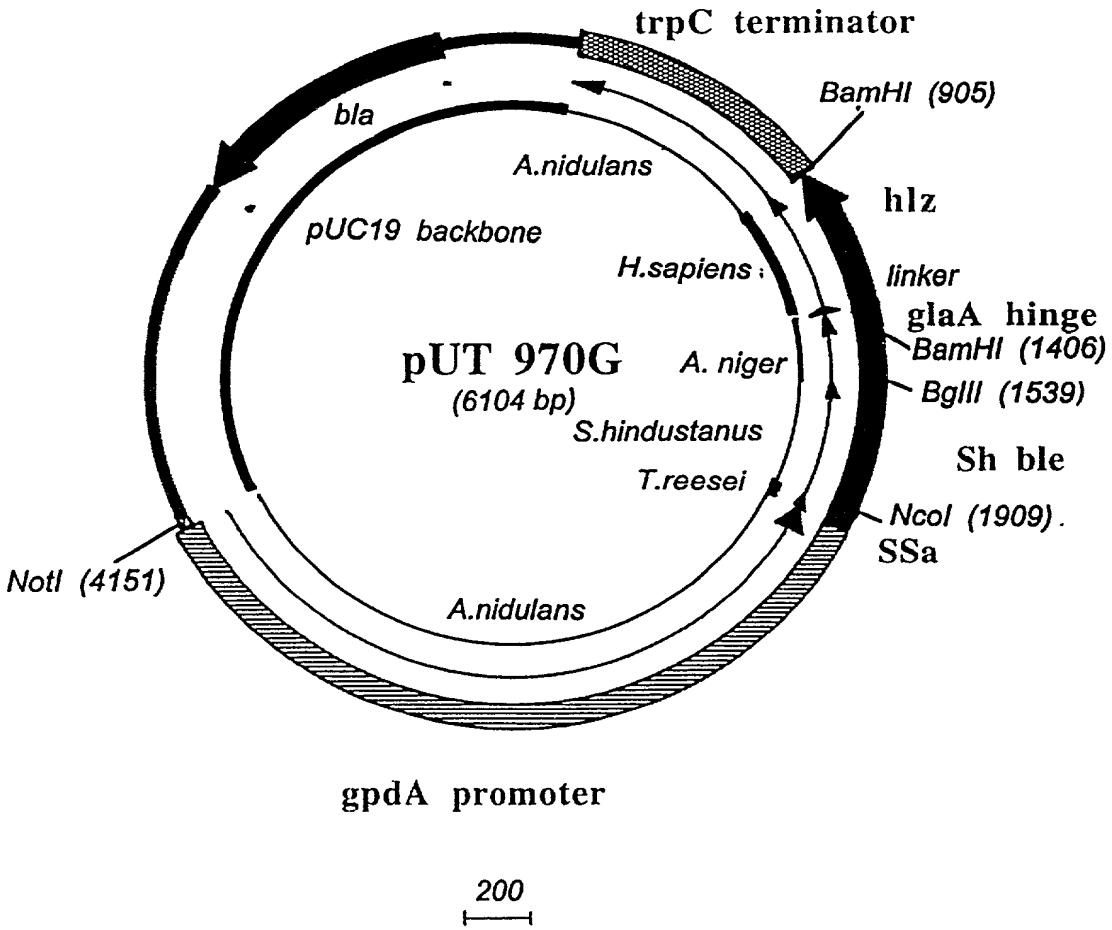

trpC terminator
BamHI (905)
bla
A.nidulans
hlz
pUC19 backbone
H.sapiens
linker
glaA hinge
pUT 970G
A. niger
BamHI (1406)
(6104 bp)
BglII (1539)
S.hindustanus
Sh ble
T.reesei
NcoI (1909)
SSa
NotI (4151)
A.nidulans
gpdA promoter
200

Fig 4

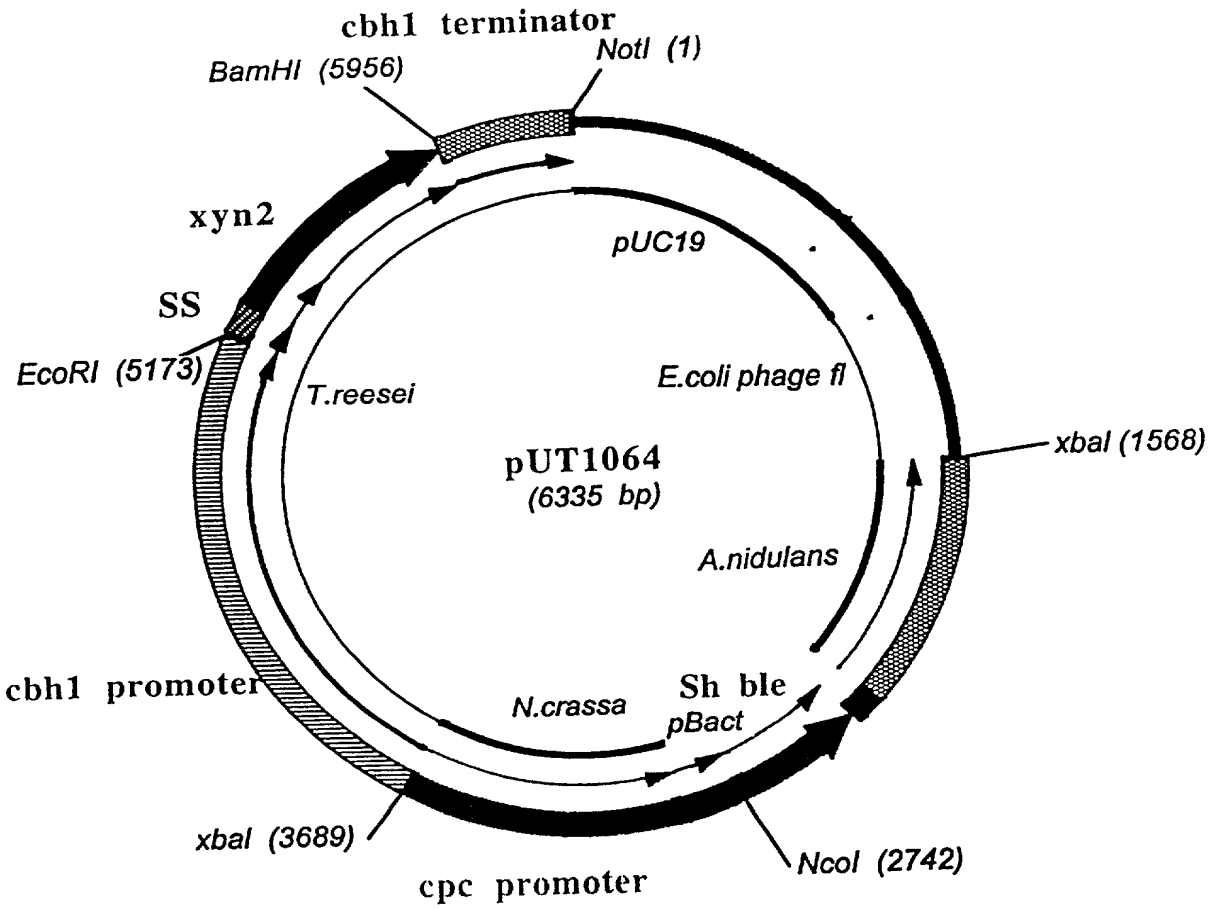

cbh1 terminator
NotI (1)
BamHI (5956)
xyn2
SS
EcoRI (5173)
pUC19
T.reesei
E.coli phage f1
pUT1064
(6335 bp)
XbaI (1568)
A.nidulans
cbh1 promoter
N.crassa
Sh ble
pBact
XbaI (3689)
NcoI (2742)
cpc promoter
200

Fig 5

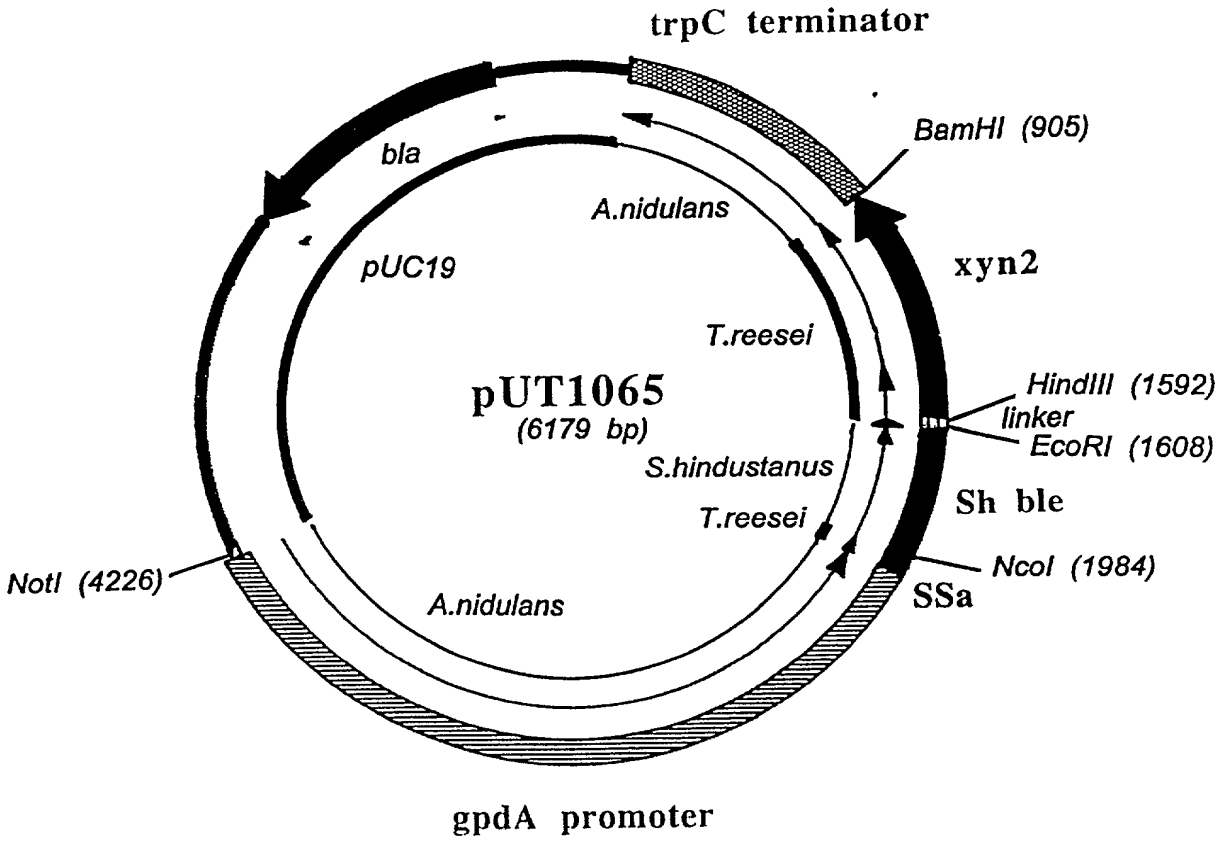

trpC terminator
BamHI (905)
bla
A.nidulans
pUC19
xyn2
T.reesei
pUT1065
(6179 bp)
HindIII (1592)
linker
EcoRI (1608)
S.hindustanus
T.reesei
Sh ble
NcoI (1984)
SSa
NotI (4226)
A.nidulans
gpdA promoter
200

Fig 6

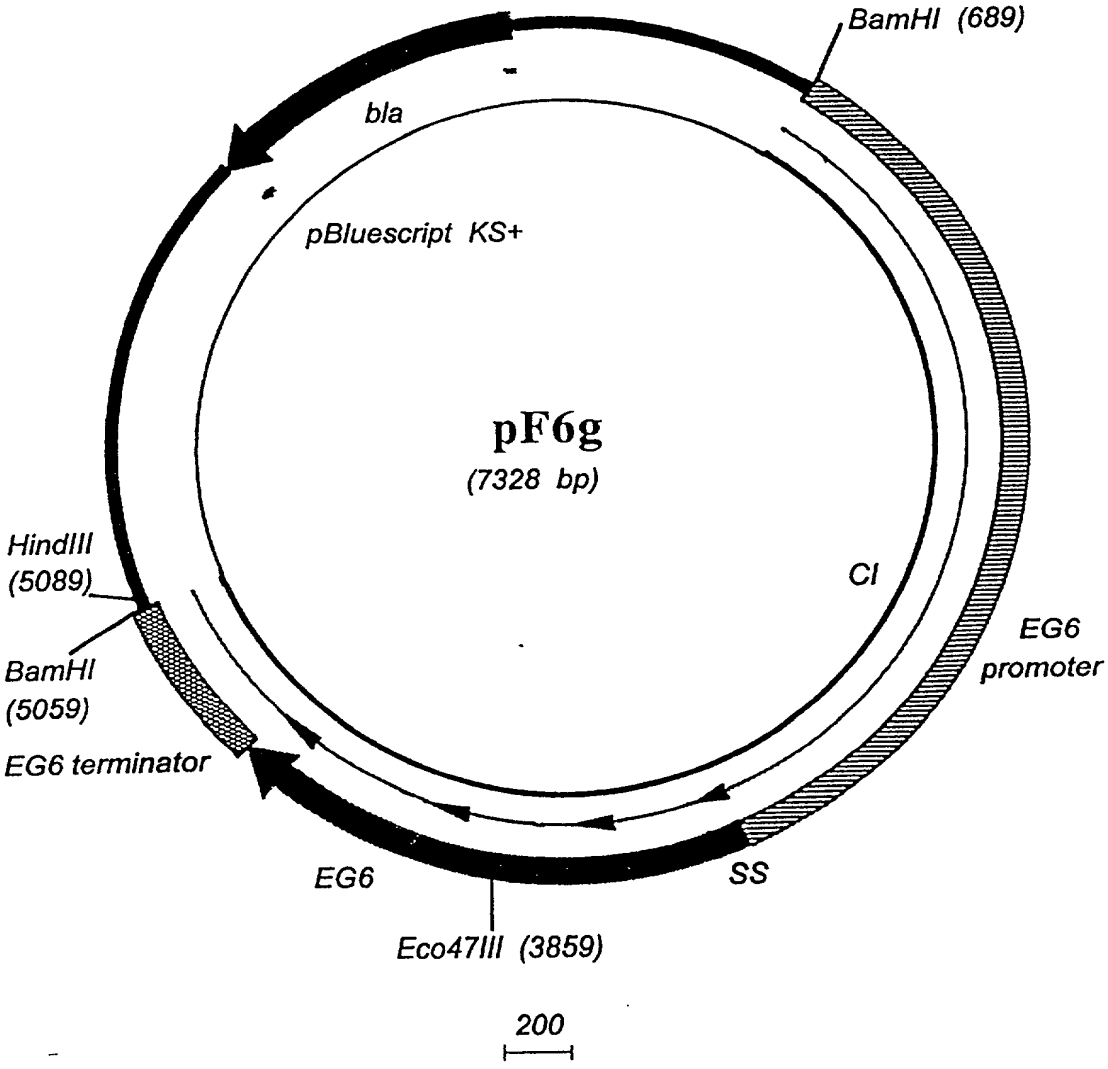
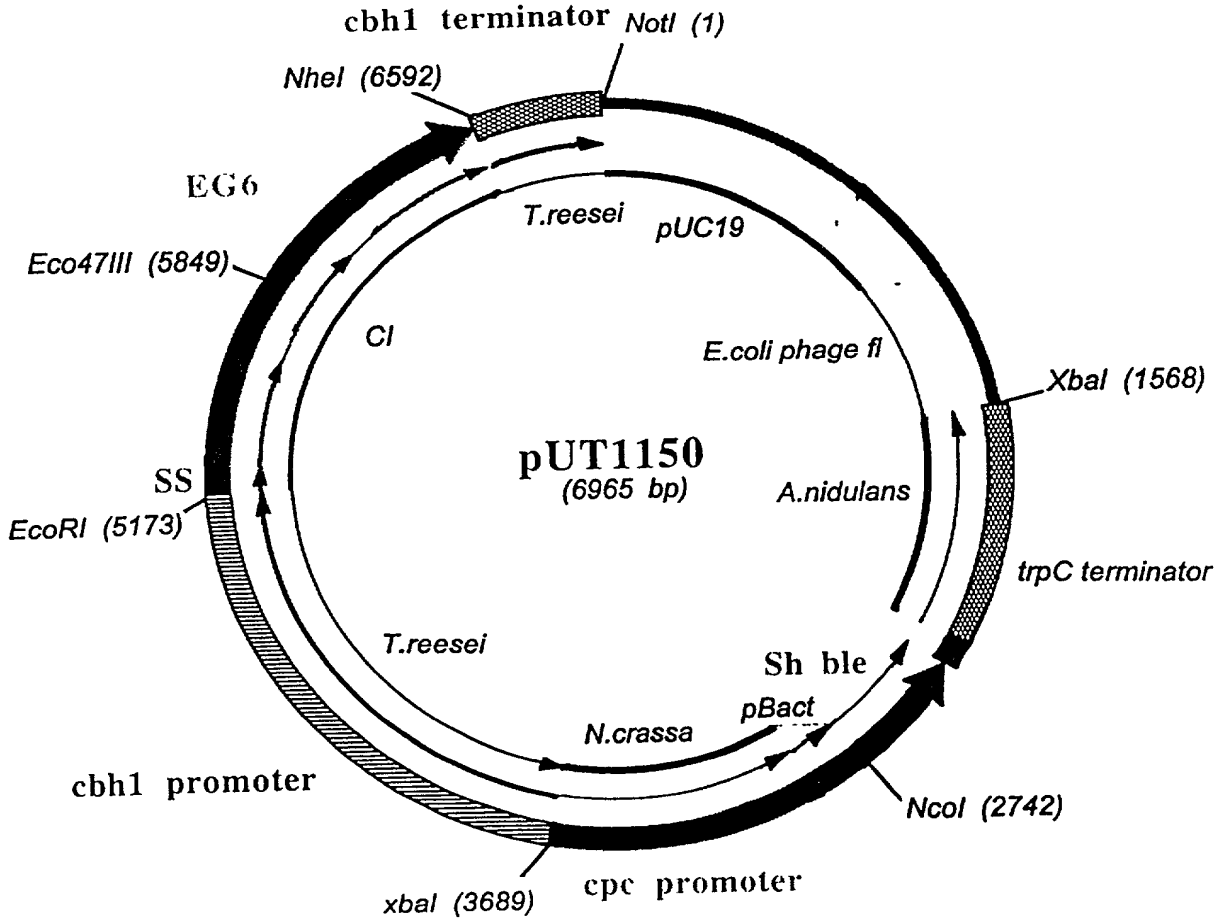

BamHI (689)
bla
pBluescript KS+
pF6g
(7328 bp)
HindIII
(5089)
BamHI
(5059)
EG6 terminator
CI
EG6
promoter
EG6
SS
Eco47III (3859)
200

Fig 7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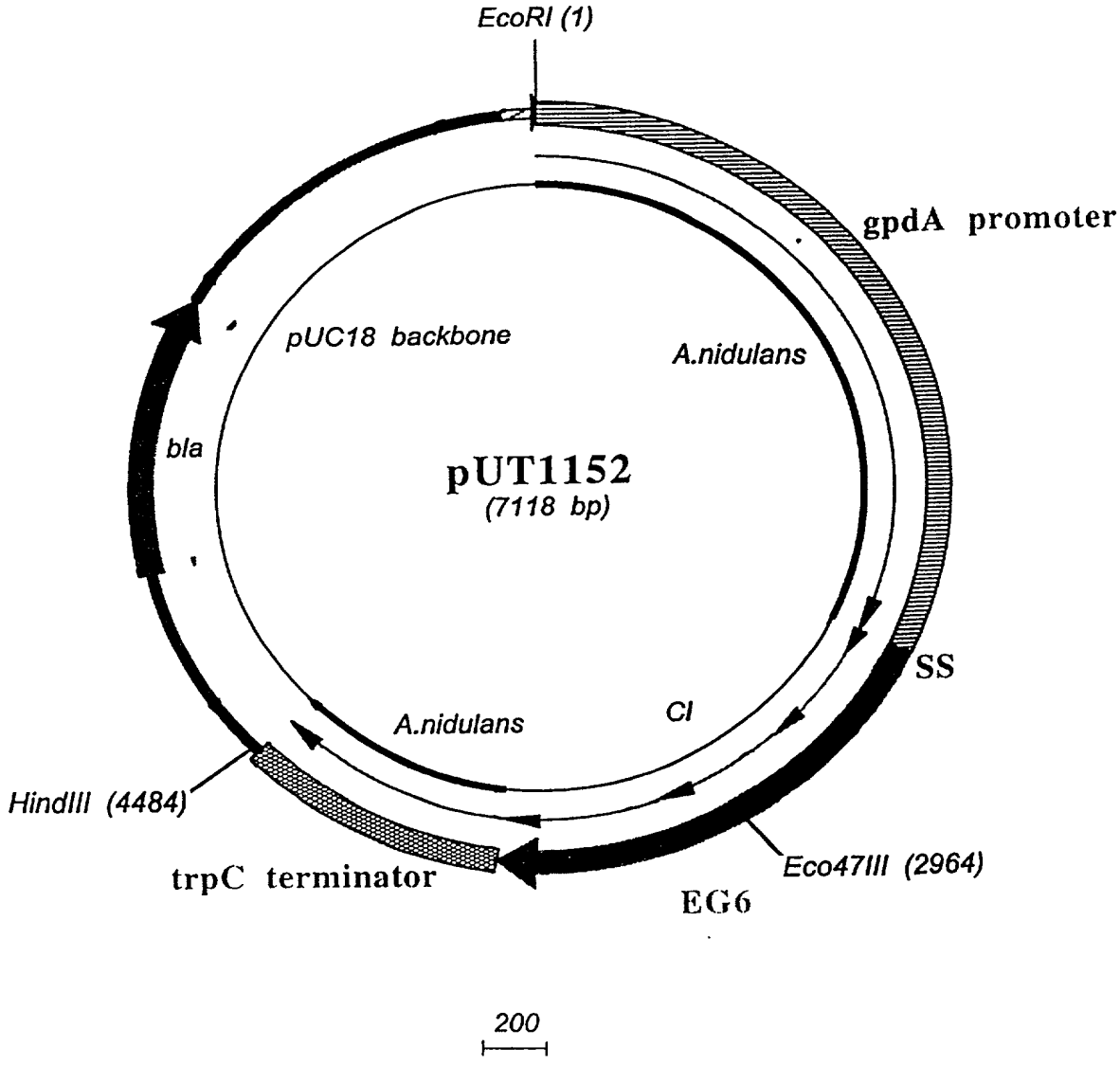
NotI (1)
NheI (6592)
EG6
T.reesei
pUC19
Eco47III (5849)
CI
E.coli phage fl
XbaI (1568)
pUT1150
(6965 bp)
SS
A.nidulans
EcoRI (5173)
trpC terminator
T.reesei
Sh ble
pBact
N.crassa
cbh1 promoter
NcoI (2742)
xbaI (3689)
cpc promoter
200

Fig 8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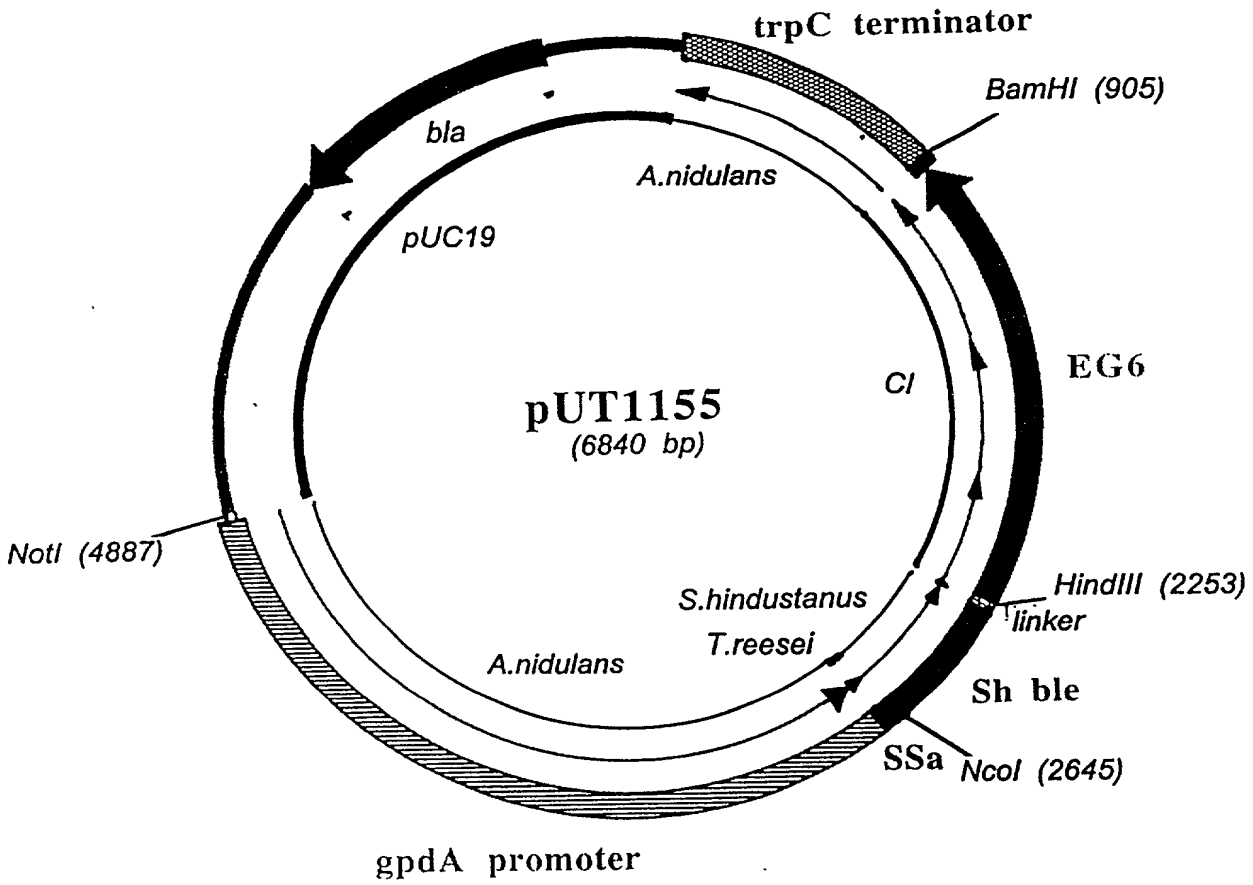
gpdA promoter
pUC18 backbone
A.nidulans
bla
pUT1152
(7118 bp)
SS
A.nidulans
CI
HindIII (4484)
Eco47III (2964)
trpC terminator
EG6
200

Fig 9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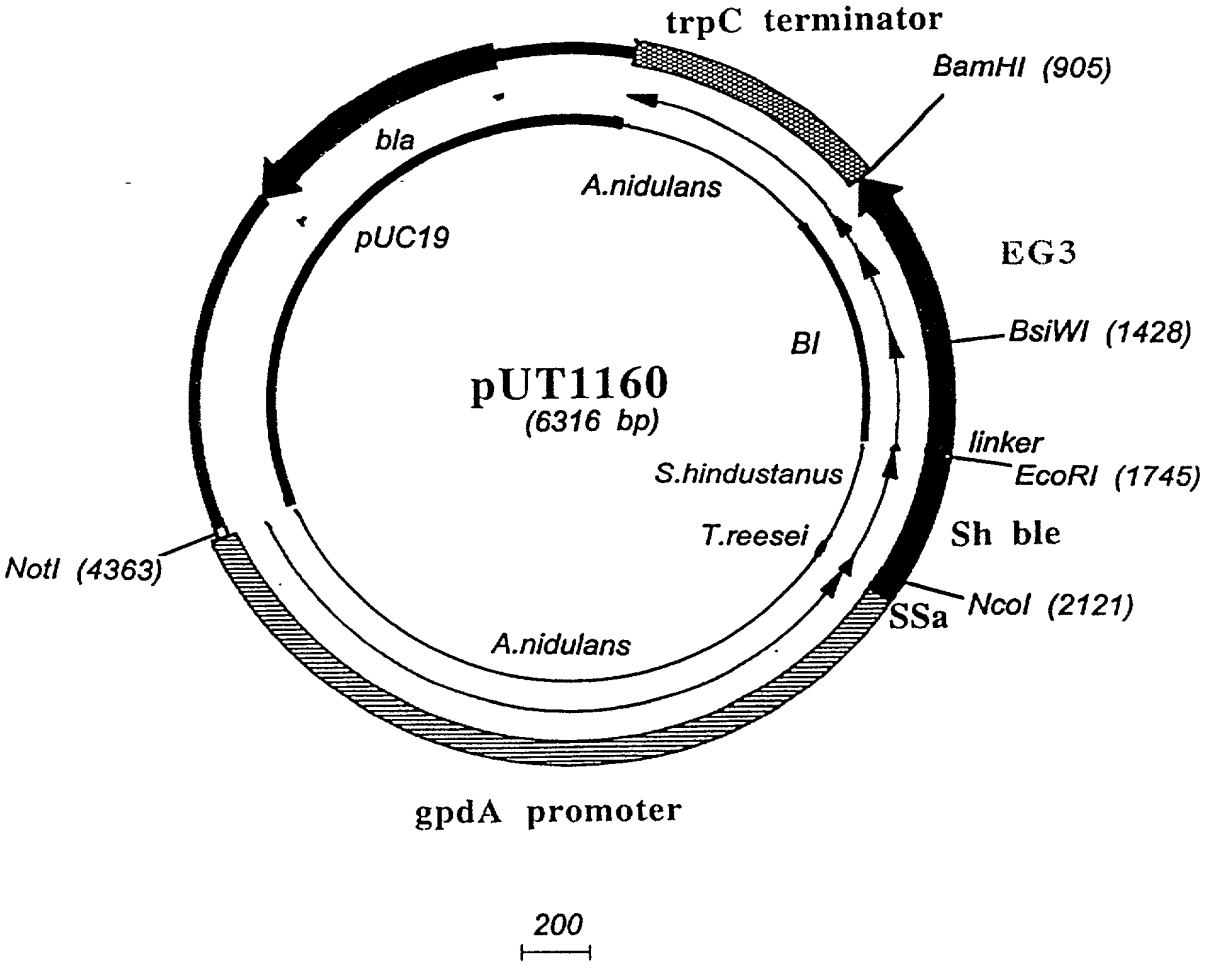
BamHI (905)
bla
A.nidulans
pUC19
pUT1155
(6840 bp)
CI
EG6
NotI (4887)
HindIII (2253)
linker
S.hindustanus
T.reesei
A.nidulans
Sh ble
SSa
NcoI (2645)
gpdA promoter
200

Fig 10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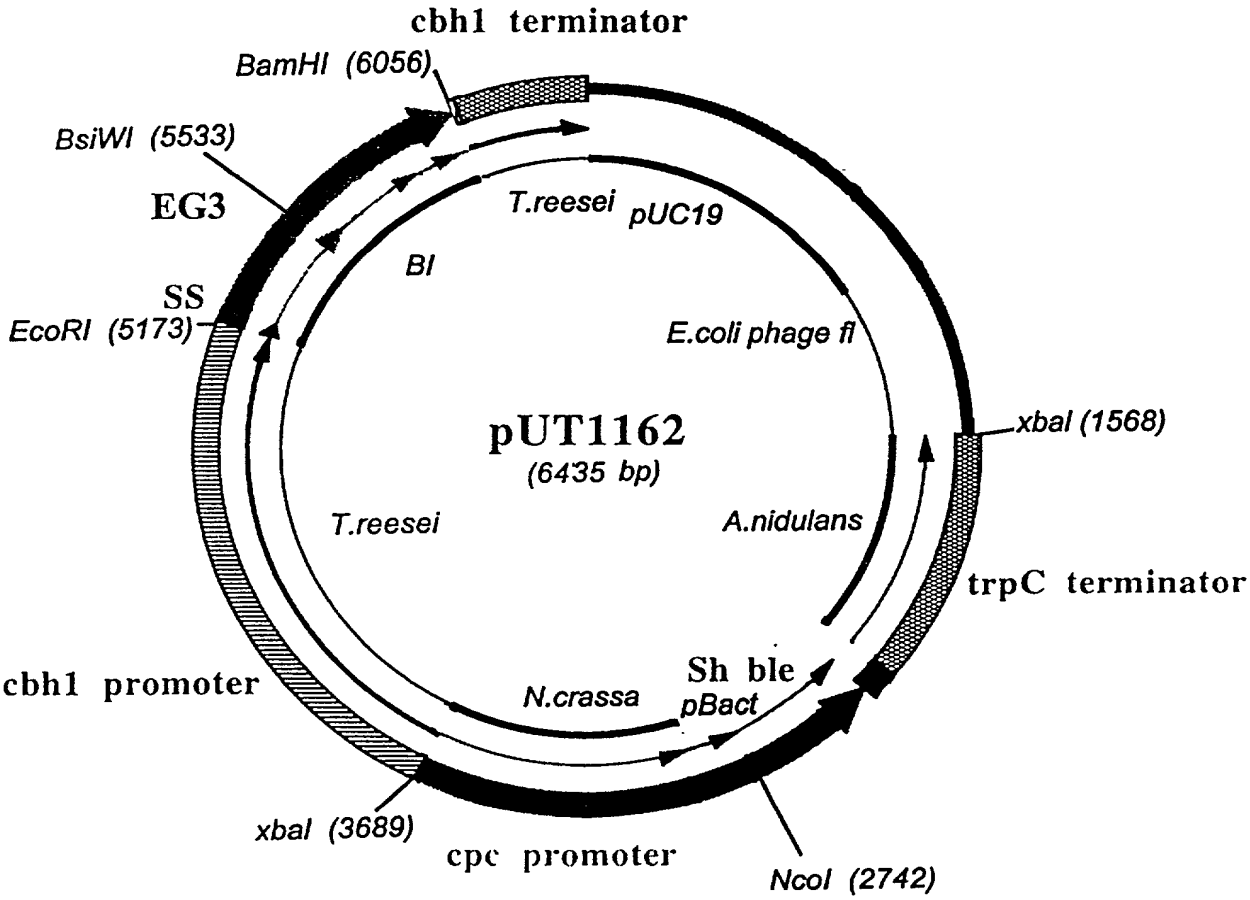
BamHI (905)
bla
A.nidulans
pUC19
EG3
BsiWI (1428)
BI
pUT1160
(6316 bp)
linker
EcoRI (1745)
S.hindustanus
T.reesei
Sh ble
NotI (4363)
NcoI (2121)
SSa
A.nidulans
gpdA promoter
200

Fig 11


cbh1 terminator
BamHI (6056)
BsiWI (5533)
EG3
SS
EcoRI (5173)
T.reesei pUC19
BI
E.coli phage fl
pUT1162
(6435 bp)
xbaI (1568)
T.reesei
A.nidulans
trpC terminator
cbh1 promoter
Sh ble
N.crassa
pBact
xbaI (3689)
cpc promoter
NcoI (2742)
200

TOETAD "4524465960

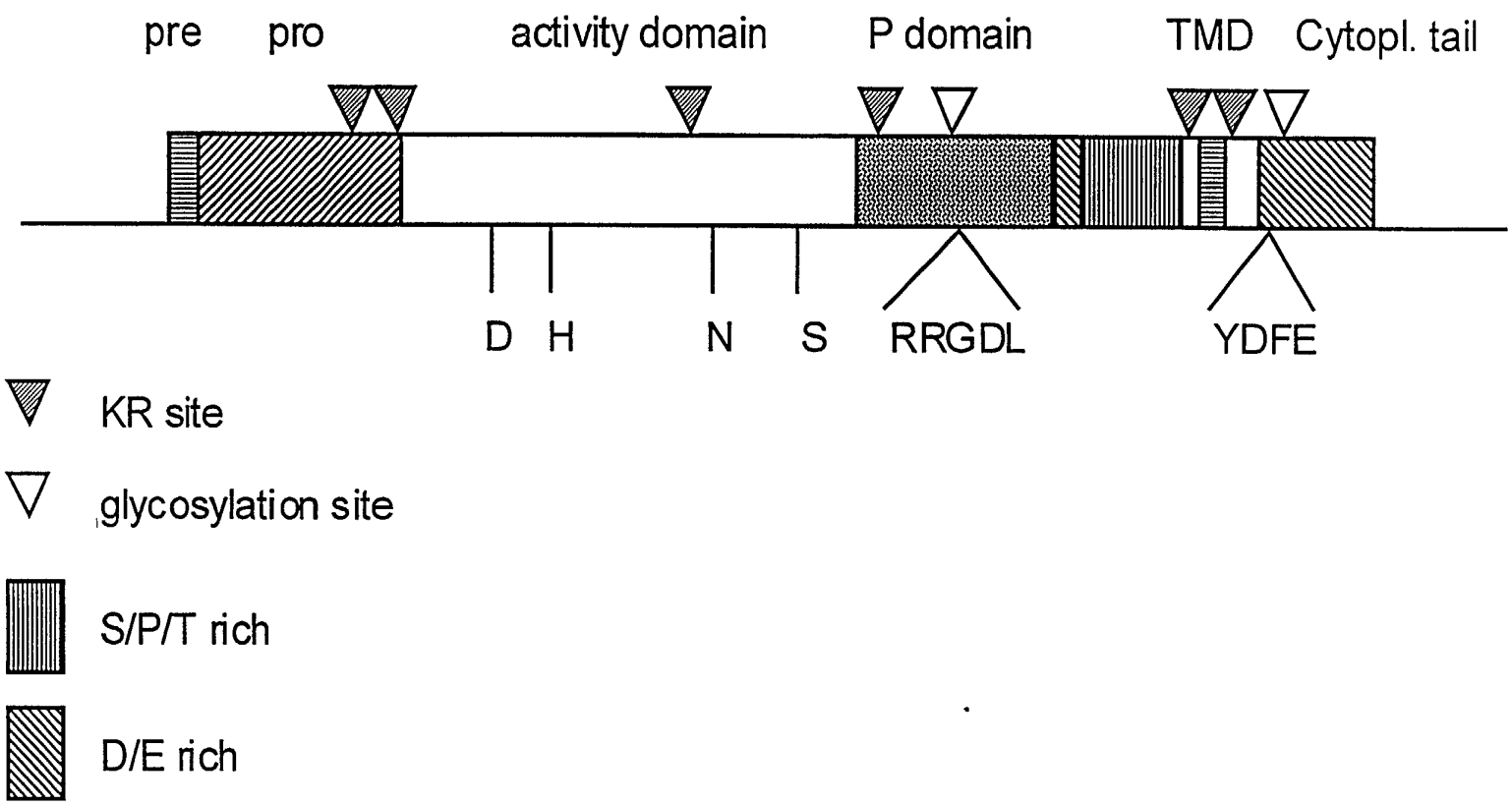


Fig 12

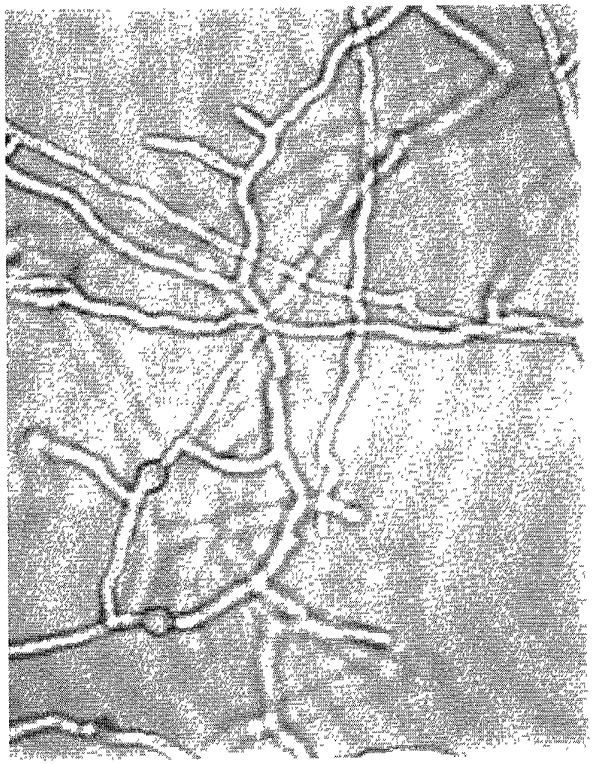


Fig 13A

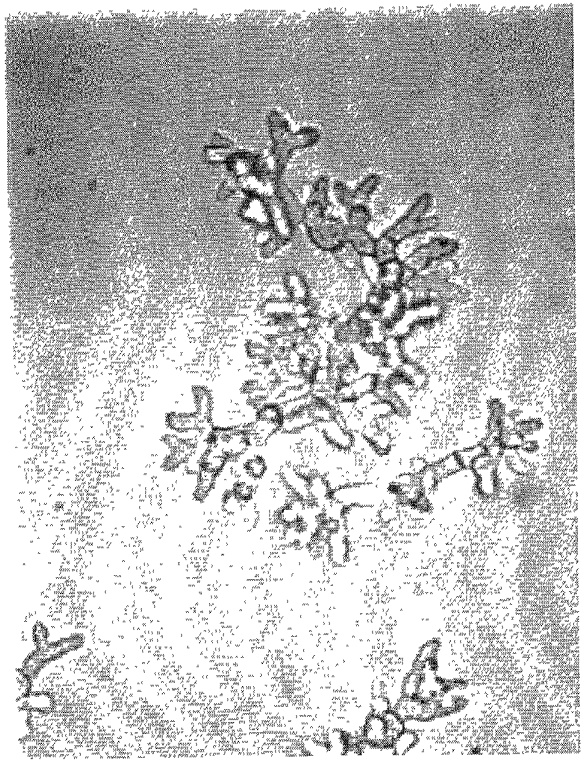


Fig 13B

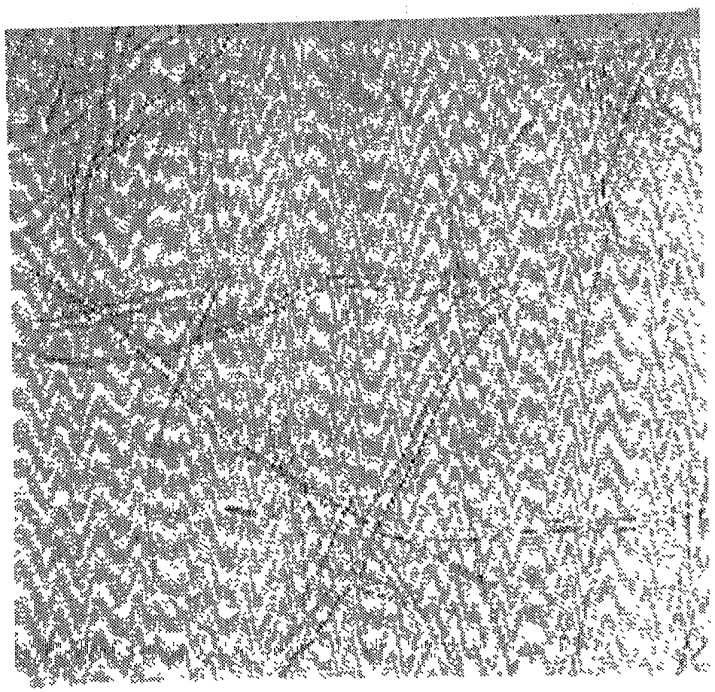


Fig. 14A

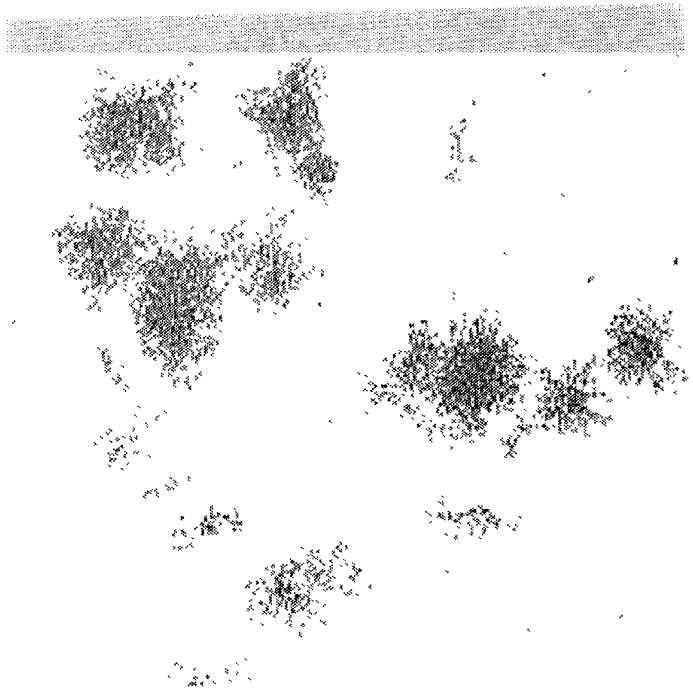


Fig. 14B

pyrE gene sequence

SEQ ID NO:1
Pyre/niger Length: 1578 March 9, 2001 09:28 Type: N Check: 2282

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  51  TTTGATTGGG TTTAATTGGA ATCTGTATAT TTTCAGATGG AGTCAACTTT
 101  TGAATGGCCA ATATATCCTC GGCGATACCG TCGGAGATAA GATAAGAATA
 151  ATCGCACACT ATTCCCAAAG CATACTGGTA CATACTGCAT TCGGCTAGTG
 201  CGGGGTGCTT ACCTCATCCA CCCGAATGAG CCCAACTTTT TTGTCTCAAT
 251  CAATAATTGC ATCCAAATTC CCCCGCAACT TCCCCCTCCA ACCCCGTGTC
 301  TATACCACTC CCTCCACACC CACACAATCA CAATGGCTCT CCCTGCCTAC
 351  AAGACCGCCT TCCTGGAGTC TCTCGTCGGC CAACGTGCTG ACTTTCGGCA
 401  CCTTCACCCT GAAGTCGGGT CGCCGTGCGT CACCCCTCCA ACACCGGCAT
 451  TATCGCAATC GGAAGACTTA CCACTGTATA CAGACTCCCC CTACTTCTTC
 501  AACGCCGGCA TCTTCAACAC CGCCTCTCTC CTCTCCGCCC TCTCCACCAT
 551  GGCCCACACC ATCATCACCT TCCTCGCTGA GAACCCTTCC ATCCCCAAGC
 601  CCGACGTCAT GCTTCGGGTA AAAAACCCCC TCTTTCCCCA ATACCCCACT
 651  TCCACTCAAC AACCCATAAA TAACTAACAA AAACCCCCTA AACAGCCCCG
 701  CATACAAAGG CATCCCCCTC GCGTGCGCCA CCCTCCTTGA ACTCAACCGC
 751  ATCGACCCCG CCACCTGGGG CAGCGTGTCC TACAGCTACA ACCGCAAAGA
 801  AGCCAAGGAT CACGGCGAAG GCGGCAACAT TGTCGGCGCC GCTCTGAAGG
 851  GCAAGACCGT GCTTGTGATC GACGATGTCA TCACGGCCGG TACCGCCATG
 901  CGTGAGACCC TCAACCTGGT CGCCAAGGAG GGCGGCAAGG TCGTCGGATT
 951  CACTGTTGCT CTGGACCGCT TGGAGAAGAT GCCCGGACCC AAGGACGAGA
1001  ACGGTGTCGA GGACGATAAG CCCAGAATGA GTGCTATGGG TCAGATCCGT
1051  AAGGAGTATG GTGTGCCCAC GACGAGTATT GTTACTCTGG ATGATTTGAT
1101  CAAGTTGATG CAGGCGAAGG GCAATGAGGC CGATATGAAG CGGTTGGAGG
1151  AGTATAGGGC TAAGTATCAG GCTAGTGATT AGTCGGTTTC ATTGACCGAT
```

FIG. 15A

```
1201  TGTTTGGGTG GGTGTGAGAG GTTAGGTTAG GTTGTGGGCG TAGGAATGAA

1251  AAGCTGTATA CATAGGGGCC TGAAGAGGTG CGTAGAGACG GTCGTGAGAT

1301  GTTTTATGTC AAAATCTTGA ACAAATGACA CCTTAAAAAA GACCCCTTGG

1351  TTTCAGCTGA ATTAGCCCGG AAAGATGCTC GGCACGCCAT GAGTCTAGCC

1401  CACTCAGTGG GCACCCGTTT CCCACATTTG AAGTGGCCGA CGCTTATTTG

1451  GCTGAGGCTG TGGCCTGGAA AGGCACTATG GCGTGCTGCG GTACAAGGCC

1501  GGGGCTGGCG TACGAACCAC GACGCCCGAA GGGAACTCTT CGGTCTTACT

1551  ACTACTATGT CCCCAGTTGA CCCCCCGA
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SEQ ID NO:2

Translation of pyrE(1-1578)
Universal code

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1          GGGTTAATGTGAAGGCGTTAGTGGTAATGTATATTAATGGTGAGATGGGCTTTGATTGGG
           CCCAATTACACTTCCGCAATCACCATTACATATAATTACCACTCTACCCGAAACTAACCC

1           G  L  M  *  R  R  *  W  *  C  I  L  M  V  R  W  A  L  I  G
1            G  *  C  E  G  V  S  G  N  V  Y  *  W  *  D  G  L  *  L  G
1             V  N  V  K  A  L  V  V  M  Y  I  N  G  E  M  G  F  D  W  V

61         TTTAATTGGAATCTGTATATTTTCAGATGGAGTCAACTTTTGAATGGCCAATATATCCTC
           AAATTAACCTTAGACATATAAAAGTCTACCTCAGTTGAAAACTTACCGGTTATATAGGAG

21          F  N  W  N  L  Y  I  F  R  W  S  Q  L  L  N  G  Q  Y  I  L
21           L  I  G  I  C  I  F  S  D  G  V  N  F  *  M  A  N  I  S  S
21            *  L  E  S  V  Y  F  Q  M  E  S  T  F  E  W  P  I  Y  P  R

121        GGCGATACCGTCGGAGATAAGATAAGAATAATCGCACACTATTCCCAAAGCATACTGGTA
           CCGCTATGGCAGCCTCTATTCTATTCTTATTAGCGTGTGATAAGGGTTTCGTATGACCAT

41          G  D  T  V  G  D  K  I  R  I  I  A  H  Y  S  Q  S  I  L  V
41           A  I  P  S  E  I  R  *  E  *  S  H  T  I  P  K  A  Y  W  Y
41            R  Y  R  R  R  *  D  K  N  N  R  T  L  F  P  K  H  T  G  T

181        CATACTGCATTCGGCTAGTGCGGGGTGCTTACCTCATCCACCCGAATGAGCCCAACTTTT
           GTATGACGTAAGCCGATCACGCCCCACGAATGGAGTAGGTGGGCTTACTCGGGTTGAAAA

61          H  T  A  F  G  *  C  G  V  L  T  S  S  T  R  M  S  P  T  F
61           I  L  H  S  A  S  A  G  C  L  P  H  P  P  E  *  A  Q  L  F
61            Y  C  I  R  L  V  R  G  A  Y  L  I  H  P  N  E  P  N  F  F
```

FIG. 15B

```
241        TTGTCTCAATCAATAATTGCATCCAAATTCCCCCGCAACTTCCCCCTCCAACCCCGTGTC
           AACAGAGTTAGTTATTAACGTAGGTTTAAGGGGGCGTTGAAGGGGGAGGTTGGGGCACAG

81          L  S  Q  S  I  I  A  S  K  F  P  R  N  F  P  L  Q  P  R  V
81           C  L  N  Q  *  L  H  P  N  S  P  A  T  S  P  S  N  P  V  S
81            V  S  I  N  N  C  I  Q  I  P  P  Q  L  P  P  P  T  P  C  L
                                                ?????????

301        TATACCACTCCCTCCACACCCACACAATCACAATGGCTCTCCCTGCCTACAAGACCGCCT
           ATATGGTGAGGGAGGTGTGGGTGTGTTAGTGTTACCGAGAGGGACGGATGTTCTGGCGGA

101         Y  T  T  P  S  T  P  T  Q  S  Q  W  L  S  L  P  T  R  P  P
101          I  P  L  P  P  H  P  H  N  H  N  G  S  P  C  L  Q  D  R  L
101           Y  H  S  L  H  T  H  T  I  T  M  A  L  P  A  Y  K  T  A  F

361        TCCTGGAGTCTCTCGTCGGCCAACGTGCTGACTTTCGGCACCTTCACCCTGAAGTCGGGT
           AGGACCTCAGAGAGCAGCCGGTTGCACGACTGAAAGCCGTGGAAGTGGGACTTCAGCCCA
            ??????????????????????????
121         S  W  S  L  S  S  A  N  V  L  T  F  G  T  F  T  L  K  S  G
121          P  G  V  S  R  R  P  T  C  *  L  S  A  P  S  P  *  S  R  V
121           L  E  S  L  V  G  Q  R  A  D  F  R  H  L  H  P  E  V  G  S

                          INTRON I
421        CGCC**GTGCGTCACCCCTCCAACACCGGCATTATCGCAATCGGAAGACTTACCACTGTATA**
           GCGGCACGCAGTGGGGAGGTTGTGGCCGTAATAGCGTTAGCCTTCTGAATGGTGACATAT

141         R  R  A  S  P  L  Q  H  R  H  Y  R  N  R  K  T  Y  H  C  I
141          A  V  R  H  P  S  N  T  G  I  I  A  I  G  R  L  T  T  V  Y
141           P  C  V  T  P  P  T  P  A  L  S  Q  S  E  D  L  P  L  Y  T

481        **CAG**ACTCCCCCTACTTCTTCAACGCCGGCATCTTCAACACCGCCTCTCTCCTCTCCGCCC
           GTCTGAGGGGGATGAAGAAGTTGCGGCCGTAGAAGTTGTGGCGGAGAGAGGAGAGGCGGG

161         Q  T  P  P  T  S  S  T  P  A  S  S  T  P  P  L  S  S  P  P
161          R  L  P  L  L  L  Q  R  R  H  L  Q  H  R  L  S  P  L  R  P
161           D  S  P  Y  F  F  N  A  G  I  F  N  T  A  S  L  L  S  A  L

                *NcoI*
541        TCTCCA*CCATGG*CCCACACCATCATCACCTTCCTCGCTGAGAACCCTTCCATCCCCAAGC
           AGAGGT*GGTACC*GGGTGTGGTAGTAGTGGAAGGAGCGACTCTTGGGAAGGTAGGGGTTCG

181         S  P  P  W  P  T  P  S  S  P  S  S  L  R  T  L  P  S  P  S
181          L  H  H  G  P  H  H  H  H  L  P  R  *  E  P  F  H  P  Q  A
181           S  T  M  A  H  T  I  I  T  F  L  A  E  N  P  S  I  P  K  P
              ??????????                    INTRON II

601        CCGACGTCATGCTTCGG**GTAAAAAACCCCCTCTTTCCCCAATACCCCACTTCCACTCAAC**
           GGCTGCAGTACGAAGCCCATTTTTTGGGGGAGAAAGGGGTTATGGGGTGAAGGTGAGTTG

201         P  T  S  C  F  G  *  K  T  P  S  F  P  N  T  P  L  P  L  N
201          R  R  H  A  S  G  K  K  P  P  L  S  P  I  P  H  F  H  S  T
201           D  V  M  L  R  V  K  N  P  L  F  P  Q  Y  P  T  S  T  Q  Q
```

FIG. 15C

661 **AACCCATAAATAACTAACAAAAACCCCCTAAACAG**CCCCGCATACAAAGGCATCCCCCTC
TTGGGTATTTATTGATTGTTTTTGGGGGATTTGTCGGGGCGTATGTTTCCGTAGGGGGAG

221 N P * I T N K N P L N S P A Y K G I P L
221 T H K * L T K T P * T A P H T K A S P S
221 P I N N * Q K P P K Q P R I Q R H P P R

721 GCGTGCGCCACCCTCCTTGAACTCAACCGCATCGACCCCGCCACCTGGGGCAGCGTGTCC
CGCACGCGGTGGGAGGAACTTGAGTTGGCGTAGCTGGGGCGGTGGACCCCGTCGCACAGG

241 A C A T L L E L N R I D P A T W G S V S
241 R A P P S L N S T A S T P P P G A A C P
241 V R H P P * T Q P H R P R H L G Q R V L

781 TACAGCTACAACCGCAAAGAAGCCAAGGATCACGGCGAAGGCGGCAACATTGTCGGCGCC
ATGTCGATGTTGGCGTTTCTTCGGTTCCTAGTGCCGCTTCCGCCGTTGTAACAGCCGCGG

261 Y S Y N R K E A K D H G E G G N I V G A
261 T A T T A K K P R I T A K A A T L S A P
261 Q L Q P Q R S Q G S R R R R Q H C R R R

KpnI
841 GCTCTGAAGGGCAAGACCGTGCTTGTGATCGACGATGTCATCACGGCC*GGTACC*GCCATG
CGAGACTTCCCGTTCTGGCACGAACACTAGCTGCTACAGTAGTGCCGG*CCATGG*CGGTAC

281 A L K G K T V L V I D D V I T A G T A M
281 L * R A R P C L * S T M S S R P V P P C
281 S E G Q D R A C D R R C H H G R Y R H A

901 CGTGAGACCCTCAACCTGGTCGCCAAGGAGGGCGGCAAGGTCGTCGGATTCACTGTTGCT
GCACTCTGGGAGTTGGACCAGCGGTTCCTCCCGCCGTTCCAGCAGCCTAAGTGACAACGA

301 R E T L N L V A K E G G K V V G F T V A
301 V R P S T W S P R R A A R S S D S L L L
301 * D P Q P G R Q G G R Q G R R I H C C S

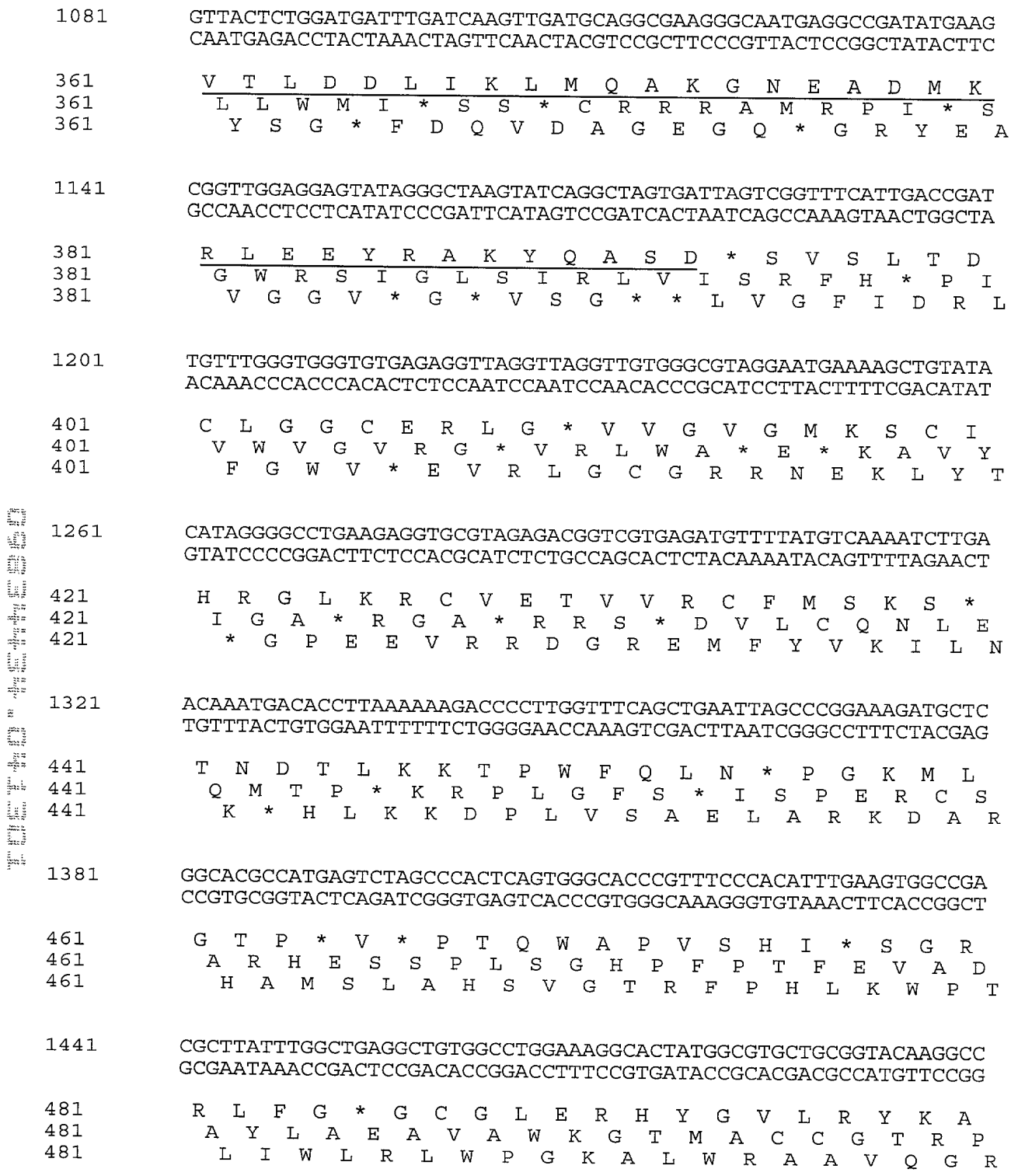
961 CTGGACCGCTTGGAGAAGATGCCCGGACCCAAGGACGAGAACGGTGTCGAGGACGATAAG
GACCTGGCGAACCTCTTCTACGGGCCTGGGTTCCTGCTCTTGCCACAGCTCCTGCTATTC

321 L D R L E K M P G P K D E N G V E D D K
321 W T A W R R C P D P R T R T V S R T I S
321 G P L G E D A R T Q G R E R C R G R * A

1021 CCCAGAATGAGTGCTATGGGTCAGATCCGTAAGGAGTATGGTGTGCCCACGACGAGTATT
GGGTCTTACTCACGATACCCAGTCTAGGCATTCCTCATACCACACGGGTGCTGCTCATAA

341 P R M S A M G Q I R K E Y G V P T T S I
341 P E * V L W V R S V R S M V C P R R V L
341 Q N E C Y G S D P * G V W C A H D E Y C

FIG. 15D


1081
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CAATGAGACCTACTAAACTAGTTCAACTACGTCCGCTTCCCGTTACTCCGGCTATACTTC
361
V T L D D L I K L M Q A K G N E A D M K
361
L L W M I * S S * C R R R A M R P I * S
361
Y S G * F D Q V D A G E G Q * G R Y E A
1141
CGGTTGGAGGAGTATAGGGCTAAGTATCAGGCTAGTGATTAGTCGGTTTCATTGACCGAT
GCCAACCTCCTCATATCCCGATTCATAGTCCGATCACTAATCAGCCAAAGTAACTGGCTA
381
R L E E Y R A K Y Q A S D * S V S L T D
381
G W R S I G L S I R L V I S R F H * P I
381
V G G V * G * V S G * * L V G F I D R L
1201
TGTTTGGGTGGGTGTGAGAGGTTAGGTTAGGTTGTGGGCGTAGGAATGAAAAGCTGTATA
ACAAACCCACCCACACTCTCCAATCCAATCCAACACCCGCATCCTTACTTTTCGACATAT
401
C L G G C E R L G * V V G V G M K S C I
401
V W V G V R G * V R L W A * E * K A V Y
401
F G W V * E V R L G C G R R N E K L Y T
1261
CATAGGGGCCTGAAGAGGTGCGTAGAGACGGTCGTGAGATGTTTTATGTCAAAATCTTGA
GTATCCCCGGACTTCTCCACGCATCTCTGCCAGCACTCTACAAAATACAGTTTTAGAACT
421
H R G L K R C V E T V V R C F M S K S *
421
I G A * R G A * R R S * D V L C Q N L E
421
* G P E E V R R D G R E M F Y V K I L N
1321
ACAAATGACACCTTAAAAAAGACCCCTTGGTTTCAGCTGAATTAGCCCGGAAAGATGCTC
TGTTTACTGTGGAATTTTTTCTGGGGAACCAAAGTCGACTTAATCGGGCCTTTCTACGAG
441
T N D T L K K T P W F Q L N * P G K M L
441
Q M T P * K R P L G F S * I S P E R C S
441
K * H L K K D P L V S A E L A R K D A R
1381
GGCACGCCATGAGTCTAGCCCACTCAGTGGGCACCCGTTTCCCACATTTGAAGTGGCCGA
CCGTGCGGTACTCAGATCGGGTGAGTCACCCGTGGGCAAAGGGTGTAAACTTCACCGGCT
461
G T P * V * P T Q W A P V S H I * S G R
461
A R H E S S P L S G H P F P T F E V A D
461
H A M S L A H S V G T R F P H L K W P T
1441
CGCTTATTTGGCTGAGGCTGTGGCCTGGAAAGGCACTATGGCGTGCTGCGGTACAAGGCC
GCGAATAAACCGACTCCGACACCGGACCTTTCCGTGATACCGCACGACGCCATGTTCCGG
481
R L F G * G C G L E R H Y G V L R Y K A
481
A Y L A E A V A W K G T M A C C G T R P
481
L I W L R L W P G K A L W R A A V Q G R

FIG. 15E

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1501      GGGGCTGGCGTACGAACCACGACGCCCGAAGGGAACTCTTCGGTCTTACTACTACTATGT
          CCCCGACCGCATGCTTGGTGCTGCGGGCTTCCCTTGAGAAGCCAGAATGATGATGATACA

501        G  A  G  V  R  T  T  T  P  E  G  N  S  S  V  L  L  L  L  C
501         G  L  A  Y  E  P  R  R  P  K  G  T  L  R  S  Y  Y  Y  Y  V
501          G  W  R  T  N  H  D  A  R  R  E  L  F  G  L  T  T  T  M  S


1561      CCCCAGTTGACCCCCCGA
          GGGGTCAACTGGGGGGCT

521        P  Q  L  T  P  R
521         P  S  *  P  P
521          P  V  D  P  P
```

FIG. 15F